CLAIMS

1. Process for the culturing of cells by continuous perfusion culturing of a cell culture comprising cell culture medium and cells, wherein:
cell culture medium is added to the cell culture,
the cell culture is circulated over a filter module comprising hollow fibers resulting in an outflow of liquid having a lower cell density than the cell culture, and the flow within the filter module is an alternating tangential flow.

2. Process according to claim 1, wherein the cell culture medium is added at a perfusion rate calculated according to Formula 1:

Perfusion rate = SPR*total cell culture volume*viable cell density (1)

wherein the perfusion rate is expressed in liters per day, wherein the SPR is the specific perfusion rate, i.e. the rate in which the cell culture medium is fed to the cell culture expressed as the volume of medium added per viable cell per time unit and wherein the viable cell density is the number of viable cells per unit of volume.

3. Process according to claim 2, wherein the SPR is between 0.01 and 0.3 nL/cell/day.

4. Process according to any one of the preceding claims, wherein biomass is removed at least once from the cell culture and additional cell culture medium is added to the cell culture.

5. Process according to claim 4, wherein the biomass removal is started just before or just after the cells have reached a steady state

6. Process according to claim 4 or 5, wherein a volume of biomass is removed of between 2 and 40% of the total volume of the cell culture per day.

7. Process according to any one of the preceding claims, wherein the alternating tangential flow is achieved using one pump to circulate the cell culture over a filter module comprising hollow fibers and using another pump to remove the

liquid having a lower cell density than the cell culture prior to the filter separation.

8. Process according to any one of the preceding claims, wherein the cells are cultured to a viable cell density of at least 80 x 10^6 cells per ml and a cell viability of at least 90%.

9. Process according to any one of the preceding claims, wherein the aggregates of at least 5 cells comprise at the most 5 % of the total amount of cells.

10. Process according to any one of the preceding claims, wherein the cells are animal cells, preferably mammalian cells, or yeast cells.

11. Process according to claim 10, wherein the cells are mammalian cells.

12. Process according to claim 11, wherein the mammalian cells are PER.C6® cells.

13. Process according to any one of the preceding claims, wherein the cells produce a biological substance.

14. Process according to claim 13, wherein the biological substance is a therapeutic or diagnostic protein, such as a monoclonal antibody, a growth factor or a peptide hormone, an enzyme, a polynucleotide, such as a viral vector used in gene therapy, or a vaccine, preferably a monoclonal antibody.

15. Process according to claim 13 or 14, wherein the biological substance is further purified in downstream processing.